

星叶草属的核形态及其系统位置*

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KARYOMORPHOLOGY AND RELATIONSHIPS OF THE GENUS *CIRCAEASTER* MAXIM.

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Abstract Investigated in this paper was the karyomorphology of the monotypic genus *Circaeaster* Maxim. The interphase nuclei and the prophase chromosomes were categorized to be simple chromocenter type and interstitial type respectively; the metaphase chromosomes were rather small, ranging from 3.00 μm to 1.20 μm in length. The karyotype was formulated as $2n = 30 = 22m + 8sm$. Its distinctive higher haploid chromosome number, together with its several highly reduced and specialized character states in morphology, suggests that *Circaeaster* is a relictual paleopolyploid group. The similar karyomorphological characteristics of interphase nuclei and prophase chromosomes between *Circaeaster* and *Kingdonia*, as well as the similar size and morphology of their metaphase chromosomes, support the viewpoint that they are closely related and are probably best treated as a ditypic family.

Key words *Circaeaster* Maxim.; Karyomorphology; Systematic position

Circaeaster, a monotypic genus which occurs in southern and southeastern Asia and has quite unique open dichotomous venation as *Kingdonia*, has been one of the most controversial genera in angiosperms in terms of its systematic position. Several families, such as the Chloranthaceae (Bentham and Hooker, 1883; Maximowicz, 1881), the Saururaceae (Junell, 1931), the Ranunculaceae (Wang, 1980; Janchen, 1949; Diels, 1932; Oliver, 1895) and the Berberidaceae (Gunderson, 1950) were once suggested to be its possible affinities. At present, the presumed relationships of *Circaeaster* with the Chloranthaceae, the Saururaceae, and the Berberidaceae have been negated (Foster, 1963). *Circaeaster* is now generally treated as a monotypic family in the Ranunculales (Berberidales) (Tamura, 1995, 1963; Wu and Kubitzki, 1993; Takhtajan, 1980, 1969; Hutchinson, 1959, 1926) or in the Circaeasterales

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(Takhtajan, 1996), or as a ditypic family which includes also *Kingdonia* (Cronquist, 1988, 1981, 1968; Thorne, 1992, 1983, 1974; Takhtajan, 1987). Very recently, the close relationship between *Circaeaster* and *Kingdonia* has been strongly supported by much evidence from embryology (Tobe, 1995), palynology (Nowicke and Skvarla, 1982), molecular data (Hoot and Crane, 1995; Oxelman and Liden, 1995) and morphology of leaf venation (Ren *et al.*, 1997; Ren and Hu, 1996), though Tamura (1995) still considered that they have no close relationship and insisted that *Kingdonia* should be retained in the Ranunculaceae while *Circaeaster* should be excluded from it and be established as a monogeneric family, the Circaeasteraceae.

Karyomorphological characters are considered to be of utmost importance for deciding systematic positions of genera in the Ranunculaceae (Tamura, 1995, 1984), but the karyotype of *Circaeaster*, except its chromosome number being reported to be $2n = 30$ (Junell, 1931), has not been reported yet. This paper is to present our observations on its karyomorphology with a view to shedding some new light on its vexed relationships.

1 Materials and Methods

Seeds of *Circaeaster agrestis* were collected from Datong County, Qinhai Province, China. The voucher specimen (Yang Qin-er 9528) was deposited in the Herbarium of Institute of Botany, Chinese Academy of Sciences (PE).

The seeds were incubated at 25°C. After germination, roots were pretreated with 0.1% colchicine for 2.5 hours, and then fixed in the 3:1 mixture of absolute ethanol and glacial acetic acid for 30 minutes. After being macerated in 1 mol/L HCl for 3~5 minutes at 60°C, they were stained with Carbol Fuchsin and then squashed.

The karyomorphological classification of interphase nuclei and prophase chromosomes followed Tanaka (1977, 1971). The karyotype formula was based on measurements of mitotic metaphase chromosomes. The symbols used to describe the karyotype followed Levan *et al.* (1964). The classification of karyotype asymmetry followed Stebbins (1971).

2 Results

In interphase nuclei (Plate 1:1), several darkly stained chromocenters were observed. The chromocenters showed irregularly protruded rough surface which gradually transformed into diffused chromatin. According to Tanaka, the interphase nuclei were categorized to be simple chromocenter type.

In prophase chromosomes (Plate 1:2), hetero- and euchromatic segments were distinguishable, but their boundaries were not clear, and the heterochromatic segments were distributed in the distal and interstitial regions as well as the proximal regions. According to Tanaka, the prophase chromosomes belonged to the interstitial type.

Metaphase chromosomes were counted to be $2n = 30$ (Plate 1:3, 4), and were rather

small in size, ranging from 3.00 μm to 1.20 μm in length (Table 1). The average length of chromosomes was 1.74 μm . The karyotype was formulated as $2n=30=22m+8sm$; no secondary constriction and satellite were detected in all the cells observed. The karyotype asymmetry was categorized as 2B type.

Table 1 Measurements of mitotic meraphase chromosomes of *Circaeaster agrestis*

Chromosome number	Length(μm)	Relative length	Arm ratio	Type	Chromosome number	Length(μm)	Relative length	Arm ratio	Type
1	$1.76 + 1.24 = 3.00$	5.76	1.42	m	16	$0.95 + 0.62 = 1.57$	3.01	1.53	sm
2	$1.71 + 1.10 = 2.81$	5.39	1.55	m	17	$0.79 + 0.77 = 1.56$	2.99	1.03	m
3	$1.64 + 0.70 = 2.34$	4.49	2.34	sm	18	$0.79 + 0.77 = 1.56$	2.99	1.03	m
4	$1.61 + 0.64 = 2.25$	4.32	2.52	sm	19	$0.93 + 0.62 = 1.55$	2.97	1.50	m
5	$1.09 + 0.93 = 2.02$	3.88	1.17	m	20	$0.93 + 0.62 = 1.55$	2.97	1.50	m
6	$1.09 + 0.93 = 2.02$	3.88	1.17	m	21	$0.93 + 0.62 = 1.55$	2.97	1.50	m
7	$1.30 + 0.65 = 1.95$	3.74	2.00	sm	22	$0.93 + 0.62 = 1.55$	2.97	1.50	m
8	$1.24 + 0.59 = 1.93$	3.70	2.10	sm	23	$0.93 + 0.62 = 1.55$	2.97	1.50	m
9	$1.09 + 0.74 = 1.83$	3.51	1.47	m	24	$0.93 + 0.62 = 1.55$	2.97	1.50	m
10	$1.09 + 0.74 = 1.83$	3.51	1.47	m	25	$1.05 + 0.43 = 1.48$	2.84	2.44	sm
11	$0.95 + 0.79 = 1.74$	3.34	1.20	m	26	$1.05 + 0.43 = 1.48$	2.84	2.44	sm
12	$0.93 + 0.78 = 1.71$	3.28	1.19	m	27	$0.74 + 0.56 = 1.30$	2.49	1.32	m
13	$1.09 + 0.53 = 1.62$	3.11	2.06	sm	28	$0.71 + 0.54 = 1.25$	2.39	1.31	m
14	$1.09 + 0.53 = 1.62$	3.11	2.06	sm	29	$0.64 + 0.56 = 1.20$	2.30	1.14	m
15	$0.95 + 0.62 = 1.57$	3.01	1.53	sm	30	$0.64 + 0.56 = 1.20$	2.30	1.14	m

3 Discussions

The basic chromosome number of $x = 15$ of *Circaeaster*, which was first reported by Junell(1935), is here confirmed. This number is unlike those of any other genera within the Ranunculaceae and other members within the order Ranunculales and, in this respect, *Circaeaster* is indeed distinctive. But this basic number seems not so hard to explain. There is no common basic number for the Ranunculaceae and the Ranunculales as a whole. In the Ranunculaceae, $x = 7$ and 8 are the most common basic numbers, while $x = 6$ and 9 also occur. As such, *Circaeaster* may well be a hypotetraploid (i.e., $4x - 2, x = 8$), similar to the European *Anemone nemorosa* (Shirreffs, 1986), but it seems more probable that the basic number of $x = 15$ of *Circaeaster* might have originated by hybridization and allopolyploidization from ancestral stocks with $x = 6$ and $x = 9$ ($6 + 9 = 15$) considering the possible close relationships between *Circaeaster* and *Kingdonia* with $x = 9$ (Tamura M N et al., 1995; Zhang, 1982). The hypothesis is strengthened to some degree by the fact that *Circaeaster* seems obviously

highly reduced and specialized in certain morphological respects. Its annual and peculiar rosette habit of growth, markedly condensed stem and inflorescence and its minute flowers all appear to be strongly derivative character states (Foster, 1963). Embryologically, *Circaeaster* is also apparently a highly specialized genus. It has orthotropous, unitegmic and tenuinucellate ovules with a small nucellus, a canal-like micropyle formed by the only elongated, thin (2~3-cell-layered) integument and *ab initio* cellular endosperm, and lacks the nucellar cap (Tobe, 1995). We consider that the high number of $2n=30$ of *Circaeaster* might be correlated with its highly reduced and specialized characters. We believe that additional cytological work, particularly molecular cytogenetic studies such as genomic *in situ* hybridization, can establish the phylogenetic basic number for *Circaeaster*, and whether or not it represents an ancient tetraploid lineage derived from aneuploidization or hybridization.

Although *Circaeaster* and *Kingdonia* have different basic chromosome numbers as mentioned above, they are karyomorphologically quite similar. Both of them have the simple chromocenter type of interphase nuclei and the interstitial type of prophase chromosomes; their mitotic metaphase chromosomes are very similar in size and morphology, ranging from $3.00\text{ }\mu\text{m}$ to $1.20\text{ }\mu\text{m}$ in length in *Circaeaster* and $3.00\text{ }\mu\text{m}$ to $1.80\text{ }\mu\text{m}$ in *Kingdonia* (Tamura M N *et al.*, 1995), and are mostly submetacentric and metacentric in both genera. Tamura M. N. *et al.* (1995) regarded the karyotype of *Kingdonia* comparable to the karyotypes of *Anemone* and *Clematis* and thus insisted that *Kingdonia* should be classified in the tribe Anemoneae of the subfamily Ranunculoideae in the Ranunculaceae. This is far from true. *Anemone* and *Clematis* have the largest chromosomes in the Ranunculaceae, with five larger pairs being metacentric and the remaining three pairs being telocentric or subtelocentric (Kurita, 1958). The karyotypes of *Anemone*, *Clematis* and their closely related genera are actually quite unique in the Ranunculaceae in terms of chromosome size and morphology. Therefore, on the basis of our present knowledge of the karyomorphology in the Ranunculaceae and in *Kingdonia*, it seems quite unlikely that *Kingdonia* is related directly to the genera of the tribe Anemoneae.

In summary, our karyomorphological results support the viewpoint that *Kingdonia* and *Circaeaster* may have close relationship. Because *Circaeaster* and *Kingdonia* share the anatomical and embryological peculiarities in common, i. e., open dichotomous leaf venation and unilacunar nodes, similar pollen with striate tectum, the small nucellus without a nucellus cap and the *ab initio* cellular type endosperm, as well as the evidence from molecular systematics, the treatment placing the two genera into a ditypic family, the Circaeasteraceae (Cronquist, 1988, 1981, 1968; Thorne, 1992, 1983, 1974; Takhtajan, 1987), is probably the most appropriate as pointed out earlier by Nowicke and Skvarla (1982).

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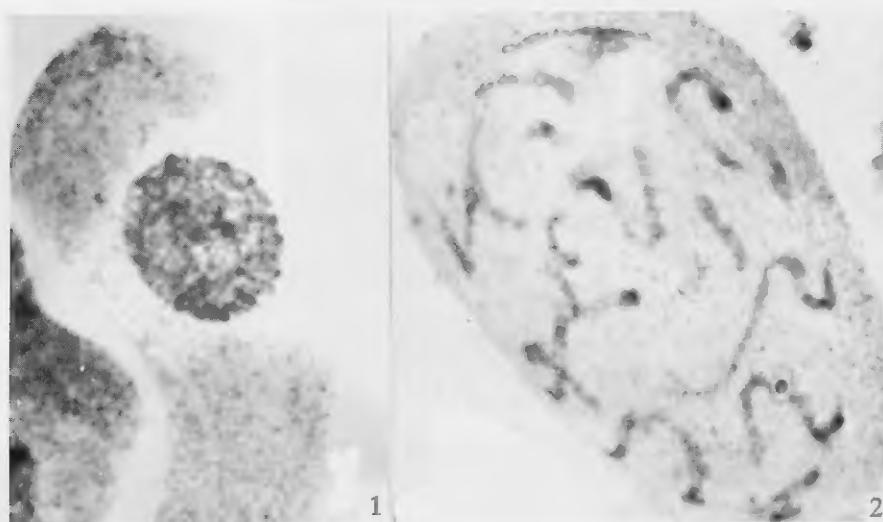
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Explanation of Plates

Plate 1 Photomicrographs of chromosomes in *Circaeaster agrestis*. 1. interphase nuclei ($\times 2000$); 2. prophase ($\times 2000$); 3. metaphase ($\times 2800$); 4. karyotype

摘要 本文研究了星叶草属的核形态。其间期核和前期染色体分别为简单染色中心型和中间型；中期染色体较小，长度介于 $3.00 \mu\text{m}$ 到 $1.20 \mu\text{m}$ 之间；核型公式为 $2n = 30 = 22m + 8sm$ 。其明显很高的染色体基数以及其它退化和特化的形态学性状，表明该属是一个孑遗的古多倍体类群。该属与独叶草属在间期核形态、前期染色体形态以及中期染色体的大小和形态方面极为相似。结合其它方面的资料，本文认为星叶草属和独叶草属有较近的亲缘关系，支持将它们一起置于星叶草科的观点。

关键词 星叶草属；核形态；系统位置



1

2



3



1

16

17

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See explanation at the end of text